FILE 'BIOSIS, MEDLINE, CAPLUS, EMBASE' ENTERED AT 15:14:26 ON 24 SEP 2002

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L1         21101 CPG
L2         19145 INTERLEUKIN 5
L3         18625 IL-5
L4         31024 INTERLEUKIN 3
L5         24076 IL-3
L6         25143 INTERLEUKIN 12
L7         23202 IL-12
L8        213043 SYNERGIS?
L9            58 L1 AND L2
L10            3 L9 AND L8
L11           32 L1 AND L4
L12            1 L11 AND L8
L13          475 L1 AND L6
L14           13 L13 AND L8
L15            9 DUP REM L14 (4 DUPLICATES REMOVED)
L16           74 L1 AND L3
L17            3 L16 AND L8
L18            1 DUP REM L17 (2 DUPLICATES REMOVED)
L19           32 L1 AND L5
L20            1 L19 AND L8
L21          467 L1 AND L7
L22           15 L21 AND L8
L23            9 DUP REM L22 (6 DUPLICATES REMOVED)

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L10 ANSWER 1 OF 3 CAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 2000:821361 CAPLUS
DOCUMENT NUMBER: 134:351996
TITLE: Intranasal immunization of mice with **CpG** DNA induces strong systemic and mucosal responses that are influenced by other mucosal adjuvants and antigen distribution
AUTHOR(S): McCluskie, Michael J.; Weeratna, Risini D.; Davis, Heather L.
CORPORATE SOURCE: Loeb Health Research Institute, Ottawa Hospital, Ottawa, K1Y 4E9, Can.
SOURCE: Molecular Medicine (New York) (2000), 6(10), 867-877
CODEN: MOMEF3; ISSN: 1076-1551
PUBLISHER: Johns Hopkins University Press
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Synthetic oligodeoxynucleotides (ODN) contg. immunostimulatory cytosine-guanine phosphate-linked dinucleotide (**CpG**) motifs are potent systemic and mucosal adjuvants in mice that have **synergistic** action with numerous other adjuvants, including alum and cholera toxin (CT). Herein, we evaluate **CpG** ODN with intranasal (IN) delivery of purified hepatitis B surface antigen (HBsAg), relative to and in combination with CT, Escherichia coli heat labile enterotoxin (LT), the B subunit of CT (CTB), and a nontoxic deriv. of LT (LTK63). BALB/c mice were immunized by IN administration of HBsAg, alone or combined with CT, LT, CTB, or LTK63, and/or **CpG** ODN, or non-**CpG** control ODN. In addn., the effect of low-or high-vol. administration was assessed, in order to target upper respiratory or entire respiratory tract, resp. HBsAg-specific systemic (Igs: IgG, IgG1, IgG2a in plasma) and mucosal (IgA in fecal, lung, vaginal, saliva, and gut samples) humoral responses, as well as cell-mediated immune responses including T-cell proliferation and cytokines (interleukins; IL-4, IL-5; interferon: IFN-.gamma.) were evaluated. **CpG** ODN, CT, and LT augmented anti-HBs titers equally, and more so than did CTB or LTK63. **CpG** ODN acted **synergistically** with CT and LT, but not CTB or LTK63 to enhance anti-HBs titers. Nevertheless, **CpG** ODN induced a more Th1-like response for all combinations, compared with the same formulation without **CpG**. Strength of induced systemic and mucosal immune responses was better with IN delivery of a large vol. A small vol. required multiple administrations and higher doses of antigen and adjuvant for equal results. This suggests that delivery of antigen to the lung and/or digestive system is superior to delivery to the nasal cavity. Our results suggest that the synergy between **CpG** ODN and native toxins (CT, LT) may depend on their enzymic activity and that the lack of synergy with nontoxic derivs. (LTB, LTK63) arises, since they do not have enzymic activity. Because both CT and LT are too toxic for use in humans, it is possible that **CpG** ODN may be combined with bacterial toxin mutants that retain some enzymic activity to optimize immune augmentation.
REFERENCE COUNT: 37 THERE ARE 37 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 2 OF 3 CAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 2000:373526 CAPLUS
DOCUMENT NUMBER: 133:129651
TITLE: Regulation of murine airway eosinophilia and Th2 cells by antigen-conjugated **CpG** oligodeoxynucleotides as a novel antigen-specific immunomodulator
AUTHOR(S): Shirota, Hidekazu; Sano, Kunio; Kikuchi, Tadashi; Tamura, Gen; Shirato, Kunio
CORPORATE SOURCE: First Department of Internal Medicine, Tohoku University School of Medicine, Sendai, 980-8574, Japan
SOURCE: Journal of Immunology (2000), 164(11), 5575-5582

CODEN: JOIMA3; ISSN: 0022-1767
PUBLISHER: American Association of Immunologists
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The characteristic features of bronchial asthma reflect the orchestrated activity of Th2 cells. Oligodeoxynucleotides contg. **CpG** motifs (**CpG**) have recently been highlighted as an immunomodulator that biases toward a Th1-dominant phenotype. We have previously reported that intratracheal coadministration of **CpG** and allergen inhibited airway eosinophilia and hyperresponsiveness in a **synergistic** manner. To substantiate the **synergism** between **CpG** and Ag, we introduced a covalently linked conjugate between **CpG** and Ag and examd. the efficacy on airway eosinophilia and Th2 cytokine prodn. We found that the conjugated form of **CpG** plus Ag was 100-fold more efficient in regulating airway eosinophilia than the unconjugated mixt. The inhibitory effects lasted for at least 2 mo. The inhibition of airway eosinophilia by the conjugate was Ag specific and assocd. with an improvement of the airway hyperresponsiveness and the unresponsiveness of the Ag-specific Th2 cells in the regional lymph nodes. The **CpG** -Ag conjugate was 100-fold more effective than the unconjugated mixt. for inducing in vitro Th1 differentiation in an IL-12-dependent manner. Our data show that **CpG** conjugated to Ag can work as a novel Ag-specific immunomodulator and imply that inhalation of allergen-**CpG** conjugate could be a desensitization therapy for patients with bronchial asthma.

REFERENCE COUNT: 48 THERE ARE 48 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L10 ANSWER 3 OF 3 CAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1999:659260 CAPLUS
DOCUMENT NUMBER: 131:285393
TITLE: Methods and products for stimulating the immune system using immunotherapeutic oligonucleotides and cytokines
INVENTOR(S): Krieg, Arthur M.; Weiner, George
PATENT ASSIGNEE(S): University of Iowa Research Foundation, USA
SOURCE: PCT Int. Appl., 91 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9951259	A2	19991014	WO 1999-US7335	19990402
WO 9951259	A3	20000113		
W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
CA 2323929	AA	19991014	CA 1999-2323929	19990402
AU 9934678	A1	19991025	AU 1999-34678	19990402
EP 1067956	A2	20010117	EP 1999-916332	19990402
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
US 6218371	B1	20010417	US 1999-286098	19990402
JP 2002510644	T2	20020409	JP 2000-542030	19990402
US 2002064515	A1	20020530	US 2001-824468	20010402
PRIORITY APPLN. INFO.:			US 1998-80729P P	19980403

US 1999-286098 A3 19990402
WO 1999-US7335 W 19990402

OTHER SOURCE(S): MARPAT 131:285393

AB The present invention relates to **synergistic** combinations of immunostimulatory **CpG** oligonucleotides and immunopotentiating cytokines. The immunopotentiating cytokine is GM-CSF, interleukin 3, **interleukin 5**, interleukin 12, interferon .gamma., TNF.alpha., Flt3 ligand or fusion protein comprising the cytokine and an antigen. The immunostimulatory **CpG** oligonucleotides and immunopotentiating cytokine are used together with antigen selected from the group consisting of a tumor antigen, microbial antigen or allergen. The antigen and the adjuvant compn. is useful for immunotherapy.

L12 ANSWER 1 OF 1 CAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1999:659260 CAPLUS
DOCUMENT NUMBER: 131:285393
TITLE: Methods and products for stimulating the immune system using immunotherapeutic oligonucleotides and cytokines
INVENTOR(S): Krieg, Arthur M.; Weiner, George
PATENT ASSIGNEE(S): University of Iowa Research Foundation, USA
SOURCE: PCT Int. Appl., 91 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9951259	A2	19991014	WO 1999-US7335	19990402
WO 9951259	A3	20000113		
W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
CA 2323929	AA	19991014	CA 1999-2323929	19990402
AU 9934678	A1	19991025	AU 1999-34678	19990402
EP 1067956	A2	20010117	EP 1999-916332	19990402
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
US 6218371	B1	20010417	US 1999-286098	19990402
JP 2002510644	T2	20020409	JP 2000-542030	19990402
US 2002064515	A1	20020530	US 2001-824468	20010402

PRIORITY APPLN. INFO.:
US 1998-80729P P 19980403
US 1999-286098 A3 19990402
WO 1999-US7335 W 19990402

OTHER SOURCE(S): MARPAT 131:285393

AB The present invention relates to **synergistic** combinations of immunostimulatory **CpG** oligonucleotides and immunopotentiating cytokines. The immunopotentiating cytokine is GM-CSF, **interleukin 3**, interleukin 5, interleukin 12, interferon .gamma., TNF.alpha., Flt3 ligand or fusion protein comprising the cytokine and an antigen. The immunostimulatory **CpG** oligonucleotides and immunopotentiating cytokine are used together with antigen selected from the group consisting of a tumor antigen, microbial antigen or allergen. The antigen and the adjuvant compn. is useful for immunotherapy.

L15 ANSWER 1 OF 9 MEDLINE DUPLICATE 1
ACCESSION NUMBER: 2001357881 MEDLINE
DOCUMENT NUMBER: 21311855 PubMed ID: 11418633
TITLE: Novel roles of **CpG** oligodeoxynucleotides as a leader for the sampling and presentation of **CpG** -tagged antigen by dendritic cells.
AUTHOR: Shirota H; Sano K; Hirasawa N; Terui T; Ohuchi K; Hattori T; Shirato K; Tamura G
CORPORATE SOURCE: First Department of Internal Medicine and Department of Dermatology, Tohoku University School of Medicine, Sendai, Japan.
SOURCE: JOURNAL OF IMMUNOLOGY, (2001 Jul 1) 167 (1) 66-74. Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200109
ENTRY DATE: Entered STN: 20010924
Last Updated on STN: 20010924
Entered Medline: 20010920

AB Oligodeoxynucleotides containing **CpG** motifs have been highlighted as potent Th1 activators. We previously reported that Ag and **CpG**, when conjugated together, **synergistically** promoted the Ag-specific Th1 development and inhibited the Th2-mediated airway eosinophilia. In this study, we examined the mechanisms underlying the **synergism** of the covalent conjugation. The **CpG**-OVA conjugate enhanced the Th1 activation and development. These characteristic features of the conjugate could not be ascribed to the polymerization of OVA, but mirrored the augmented binding of the **CpG**-tagged Ag to dendritic cells (DCs) in a **CpG**-guided manner, because phycobiliprotein, R-PE, conjugated to **CpG** stained a higher proportion of DCs with higher intensity than the mixture. R-PE fluorescence was emitted from cytoplasmic portions of the DCs, which simultaneously expressed costimulatory molecules and IL-12. The **CpG**-conjugated R-PE trafficking described above actually served as a potent Ag. These results indicate that **CpG** conjugated to Ag exhibit novel joint properties as promoters of Ag uptake and DC activators, thereby potentiating the ability of DCs to generate Th1 cells. The DNA-mediated promotion of Ag uptake would be advantageous for evoking host immune responses against invading microorganisms.

L15 ANSWER 2 OF 9 MEDLINE DUPLICATE 2
ACCESSION NUMBER: 2000281654 MEDLINE
DOCUMENT NUMBER: 20281654 PubMed ID: 10820244
TITLE: **CpG** oligonucleotides are potent adjuvants for the activation of autoreactive encephalitogenic T cells in vivo.
AUTHOR: Segal B M; Chang J T; Shevach E M
CORPORATE SOURCE: Laboratory of Immunology, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Bethesda, MD 20892, USA.
SOURCE: JOURNAL OF IMMUNOLOGY, (2000 Jun 1) 164 (11) 5683-8. Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals; AIDS
ENTRY MONTH: 200006
ENTRY DATE: Entered STN: 20000629
Last Updated on STN: 20000629
Entered Medline: 20000621

AB The mechanism of action of microbial adjuvants in promoting the differentiation of autoimmune effector cells remains to be elucidated. We

demonstrate that **CpG**-containing oligodeoxynucleotides (ODN) can completely substitute for heat-killed mycobacteria in the priming of encephalitogenic myelin-reactive T cells in vivo. The adjuvanticity of the **CpG** ODN was secondary to their direct ability to induce IL-12 or to act **synergistically** with endogenous IL-12 to promote Th1 differentiation and encephalitogenicity. T cells primed in the absence of **CpG** with Ag and IFA alone appeared to be in a transitional state and had not undergone differentiation along a conventional Th pathway. Unlike Th2 cells, they expressed low levels of the IL-12R beta 2 subunit and retained the ability to differentiate into encephalitogenic effectors when reactivated in vitro under Th1-polarizing conditions. These results support the use of **CpG** ODN as adjuvants but also suggest that they could potentially trigger autoimmune disease in a susceptible individual.

L15 ANSWER 3 OF 9 MEDLINE
ACCESSION NUMBER: 2002263872 MEDLINE
DOCUMENT NUMBER: 21990298 PubMed ID: 11994440
TITLE: IFN-alpha beta promote priming of antigen-specific CD8+ and CD4+ T lymphocytes by immunostimulatory DNA-based vaccines.
AUTHOR: Cho Hearn Jay; Hayashi Tomoko; Datta Sandip K; Takabayashi Kenji; Van Uden John Henry; Horner Anthony; Corr Maripat; Raz Eyal
CORPORATE SOURCE: Division of Hematology/Medical Oncology, Department of Medicine, New York Presbyterian Hospital and Cornell Medical Center, 525 East 68th Street, New York, NY 10021.. hjc2001@med.cornell.edu
CONTRACT NUMBER: AI 40682 (NIAID)
AI 47078 (NIAID)
AR 44850 (NIAMS)
SOURCE: JOURNAL OF IMMUNOLOGY, (2002 May 15) 168 (10) 4907-13. Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200206
ENTRY DATE: Entered STN: 20020511
Last Updated on STN: 20020620
Entered Medline: 20020619

AB Immunostimulatory sequence (ISS) DNA containing unmethylated **CpG** dinucleotides stimulate NK and APC to secrete proinflammatory cytokines, including IFN-alphabeta and -gamma, TNF-alpha, and IL-6 and -12, and to express costimulatory surface molecules such as CD40, B7-1, and B7-2. Although ISS DNA has little direct effect on T cells by these criteria, immunization of wild-type mice with ISS DNA and OVA results in Ag-specific CTL and Th1-type T helper activity. This investigation examines the mechanisms by which ISS DNA primes CD8(+) and CD4(+) lymphocyte activities. In this report we demonstrate that ISS DNA regulates the expression of costimulatory molecules and TAP via a novel autocrine or paracrine IFN-alphabeta pathway. Coordinated regulation of B7 costimulation and TAP-dependent cross-presentation results in priming of Ag-specific CD8(+) CTL, whereas CD40, B7, and IL-12 costimulation is required for priming of CD4(+) Th cells by ISS-based vaccines.

L15 ANSWER 4 OF 9 MEDLINE
ACCESSION NUMBER: 2001544514 MEDLINE
DOCUMENT NUMBER: 21475558 PubMed ID: 11592079
TITLE: Toll-like receptor expression reveals **CpG** DNA as a unique microbial stimulus for plasmacytoid dendritic cells which synergizes with CD40 ligand to induce high amounts of IL-12.
AUTHOR: Krug A; Towarowski A; Britsch S; Rothenfusser S; Hornung V; Bals R; Giese T; Engelmann H; Endres S; Krieg A M; Hartmann

G

CORPORATE SOURCE: Department of Internal Medicine and Division of Clinical Pharmacology, University of Munich, Munich, Germany.

SOURCE: EUROPEAN JOURNAL OF IMMUNOLOGY, (2001 Oct) 31 (10) 3026-37.
Journal code: 1273201. ISSN: 0014-2980.
PUB. COUNTRY: Germany: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200112
ENTRY DATE: Entered STN: 20011010
Last Updated on STN: 20020122
Entered Medline: 20011204

AB Human plasmacytoid dendritic cells (DC) (PDC, CD123+) and myeloid DC (MDC, CD11c+) may be able to discriminate between distinct classes of microbial molecules based on a different pattern of Toll-like receptor (TLR) expression. TLR1-TLR9 were examined in purified PDC and MDC. TLR9, which is critically involved in the recognition of **CpG** motifs in mice, was present in PDC but not in MDC. TLR4, which is required for the response to LPS, was selectively expressed on MDC. Consistent with TLR expression, PDC were susceptible to stimulation by **CpG** oligodeoxynucleotide (ODN) but not by LPS, while MDC responded to LPS but not to **CpG** ODN. In PDC, **CpG** ODN supported survival, activation (CD80, CD86, CD40, MHC class II), chemokine production (IL-8, IP-10) and maturation (CD83). CD40 ligand (CD40L) and **CpG** ODN synergized to activate PDC and to stimulate the production of IFN-alpha and IL-12 including bioactive IL-12 p70. Previous incubation of PDC with IL-3 decreased the amount of **CpG**-induced IFN-alpha and shifted the cytokine response in favor of IL-12. **CpG** ODN-activated PDC showed an increased ability to stimulate proliferation of naive allogeneic CD4 T cells, butTh1 polarization of developing T cells required simultaneous activation of PDC by CD40 ligation and **CpG** ODN. **CpG** ODN-stimulated PDC expressed CCR7, which mediates homing to lymph nodes. In conclusion, our studies reveal that IL-12 p70 production by PDC is under strict control of two signals, an adequate exogenous microbial stimulus such as **CpG** ODN, and CD40L provided endogenously by activated T cells. Thus, **CpG** ODN acts as an enhancer of T cell help, while T cell-controlled restriction to foreign antigens is maintained.

L15 ANSWER 5 OF 9 MEDLINE

ACCESSION NUMBER: 2001470539 MEDLINE
DOCUMENT NUMBER: 21406644 PubMed ID: 11515823

TITLE: Bacterial DNA does not increase serum corticosterone concentration or prevent increases induced by other stimuli.

AUTHOR: Myers L P; Krieg A M; Pruett S B
CORPORATE SOURCE: Department of Cellular Biology and Anatomy, Louisiana State University Health Sciences Center, Shreveport 71130, USA.

CONTRACT NUMBER: AA09505 (NIAAA)
CA66570 (NCI)
DK25295 (NIDDK)
DK54759 (NIDDK)
ES09158 (NIEHS)
+
SOURCE: Int Immunopharmacol, (2001 Aug) 1 (8) 1605-14.
Journal code: 100965259. ISSN: 1567-5769.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200201
ENTRY DATE: Entered STN: 20010823
Last Updated on STN: 20020125

Entered Medline: 20020122

AB Bacterial DNA containing unmethylated **CpG** motifs (**CpG** DNA) and other microbial molecules such as lipopolysaccharide (LPS) have a broad range of immune stimulatory effects, which may include many shared cell signaling pathways leading to enhanced cytokine production. Some cytokines activate the hypothalamic-pituitary-adrenal (HPA) axis, and their production is downregulated by products of the HPA axis (glucocorticoids). Because such interactions have practical implications in the clinical use of **CpG** DNA, the present study was done to examine the effects of **CpG** DNA and LPS on serum corticosterone concentrations. In contrast to LPS, administration of **CpG** DNA (DNA from Escherichia coli) (30-300 microg) alone did not significantly increase serum corticosterone concentrations 1 or 4 h after administration. Administration of **CpG** DNA to mice prior to LPS caused a **synergistic** increase in serum tumor necrosis factor-alpha (TNF-alpha), indicative of an immune stimulatory effect. LPS and TNF-alpha, however, induced similar levels of corticosterone with or without concomitant **CpG** DNA. Increasing doses of LPS caused peak corticosterone levels similar to those induced by LPS in combination with **CpG** DNA. Exogenous TNF-alpha administered in vivo induced comparable concentrations of corticosterone with or without **CpG** DNA. An alternative stressor (restraint) yielded similar levels of corticosterone with or without **CpG** DNA. These results indicate that **CpG** DNA does not induce corticosterone release or alter its release by other stimuli, indicating biologically important differences in its immune effect compared to those of LPS, and possibly reduced toxicity.

L15 ANSWER 6 OF 9 MEDLINE
ACCESSION NUMBER: 2000155542 MEDLINE
DOCUMENT NUMBER: 20155542 PubMed ID: 10693875
TITLE: The features of arthritis induced by **CpG** motifs in bacterial DNA.
AUTHOR: Deng G M; Tarkowski A
CORPORATE SOURCE: Department of Rheumatology, University of Goteborg, Sweden.
SOURCE: ARTHRITIS AND RHEUMATISM, (2000 Feb) 43 (2) 356-64. Journal code: 0370605. ISSN: 0004-3591.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200003
ENTRY DATE: Entered STN: 20000320
Last Updated on STN: 20000320
Entered Medline: 20000309

AB OBJECTIVE: To investigate the features of arthritis induced by bacterial DNA that contain **CpG** motifs. METHODS: Bacterial DNA originating from Escherichia coli and Staphylococcus aureus or synthetic oligonucleotides containing **CpG** motifs were injected directly into knee joints of mice. Histopathologic joint damage, antibody levels, cytokine levels, and synovial messenger RNA (mRNA) expression of cytokines and chemokines were assessed. RESULTS: Histopathologic signs of arthritis were evident within 2 hours and lasted for at least 3 weeks. Nonmethylated **CpG** motifs were responsible for the induction of arthritis since oligonucleotides containing these motifs triggered arthritis, whereas methylation of these nucleotides abrogated the inflammatory response. Arthritis was characterized by an influx of monocytic, Mac-1+ cells and by a scarcity of T lymphocytes. The disease was characterized locally by mRNA expression of macrophage-derived cytokines (tumor necrosis factor alpha, **interleukin-12** [IL-12], IL-1beta) and chemokines (monocyte chemoattractant protein 1, RANTES) in arthritic joints. Systemically, the arthritis was characterized by increased levels of circulating IL-6 and immunoglobulins. CONCLUSION: These findings demonstrate that bacterial DNA that contain nonmethylated **CpG** motifs induces arthritis, suggesting an important pathogenic role for

bacterial DNA in septic arthritis.

L15 ANSWER 7 OF 9 MEDLINE
ACCESSION NUMBER: 1998451440 MEDLINE
DOCUMENT NUMBER: 98451440 PubMed ID: 9780160
TITLE: Cyclosporin A enhances IL-12 production by **CpG** motifs in bacterial DNA and synthetic oligodeoxynucleotides.
AUTHOR: Redford T W; Yi A K; Ward C T; Krieg A M
CORPORATE SOURCE: University of Iowa College of Pharmacy, Iowa City 52242, USA.
CONTRACT NUMBER: DK25295 (NIDDK)
P01CA665078 (NCI)
R29-AR42556 (NIAMS)
SOURCE: JOURNAL OF IMMUNOLOGY, (1998 Oct 15) 161 (8) 3930-5.
Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199811
ENTRY DATE: Entered STN: 19990106
Last Updated on STN: 19990106
Entered Medline: 19981104

AB Certain sequences of nucleotides (**CpG** motifs) in bacterial DNA or synthetic oligonucleotides (**CpG** DNA) promote the production of proinflammatory cytokines, including TNF-alpha, IFN-gamma, IL-6, and IL-12. Here we demonstrate that the immunosuppressant cyclosporin A (CsA) unexpectedly enhanced **CpG** DNA-induced IL-12 production in murine splenocytes. CsA did not inhibit **CpG** DNA-induced TNF-alpha or IL-6 production, but decreased the production of IFN-gamma by **CpG** DNA. Upon examining mechanisms by which CsA increases IL-12 production, we found that **CpG** DNA can also induce IL-10 production in B cells and that this production was sensitive to CsA. IL-10 has anti-inflammatory effects and can reduce the production of IL-12. To determine the possible role of CsA-modulated IL-10 production in mediating the increased IL-12 levels, splenocytes from IL-10 gene-disrupted mice (IL-10 -/-) and splenocytes cultured in anti-IL-10 Ab were studied. **CpG** DNA-stimulated IL-10 (-/-) splenocytes demonstrated no increase in IL-12 levels in the presence of CsA. Anti-IL-10 Ab treatment of normal splenocytes increased the magnitude of **CpG** DNA-induced IL-12 production to that seen with CsA. These results suggest that **CpG** DNA induces CsA-sensitive IL-10 production in B cells and that IL-10 acts as a negative feedback regulator of **CpG** DNA-induced IL-12 production.

L15 ANSWER 8 OF 9 CAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 2000:373526 CAPLUS
DOCUMENT NUMBER: 133:129651
TITLE: Regulation of murine airway eosinophilia and Th2 cells by antigen-conjugated **CpG** oligodeoxynucleotides as a novel antigen-specific immunomodulator
AUTHOR(S): Shirota, Hidekazu; Sano, Kunio; Kikuchi, Tadashi; Tamura, Gen; Shirato, Kunio
CORPORATE SOURCE: First Department of Internal Medicine, Tohoku University School of Medicine, Sendai, 980-8574, Japan
SOURCE: Journal of Immunology (2000), 164(11), 5575-5582
CODEN: JOIMA3; ISSN: 0022-1767
PUBLISHER: American Association of Immunologists
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The characteristic features of bronchial asthma reflect the orchestrated activity of Th2 cells. Oligodeoxynucleotides contg. **CpG** motifs

(**CpG**) have recently been highlighted as an immunomodulator that biases toward a Th1-dominant phenotype. We have previously reported that intratracheal coadministration of **CpG** and allergen inhibited airway eosinophilia and hyperresponsiveness in a **synergistic** manner. To substantiate the **synergism** between **CpG** and Ag, we introduced a covalently linked conjugate between **CpG** and Ag and examd. the efficacy on airway eosinophilia and Th2 cytokine prodn. We found that the conjugated form of **CpG** plus Ag was 100-fold more efficient in regulating airway eosinophilia than the unconjugated mixt. The inhibitory effects lasted for at least 2 mo. The inhibition of airway eosinophilia by the conjugate was Ag specific and assocd. with an improvement of the airway hyperresponsiveness and the unresponsiveness of the Ag-specific Th2 cells in the regional lymph nodes. The **CpG** -Ag conjugate was 100-fold more effective than the unconjugated mixt. for inducing in vitro Th1 differentiation in an IL-12-dependent manner. Our data show that **CpG** conjugated to Ag can work as a novel Ag-specific immunomodulator and imply that inhalation of allergen-**CpG** conjugate could be a desensitization therapy for patients with bronchial asthma.

REFERENCE COUNT: 48 THERE ARE 48 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L15 ANSWER 9 OF 9 CAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1999:659260 CAPLUS
DOCUMENT NUMBER: 131:285393
TITLE: Methods and products for stimulating the immune system using immunotherapeutic oligonucleotides and cytokines
INVENTOR(S): Krieg, Arthur M.; Weiner, George
PATENT ASSIGNEE(S): University of Iowa Research Foundation, USA
SOURCE: PCT Int. Appl., 91 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9951259	A2	19991014	WO 1999-US7335	19990402
WO 9951259	A3	20000113		
W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
CA 2323929	AA	19991014	CA 1999-2323929	19990402
AU 9934678	A1	19991025	AU 1999-34678	19990402
EP 1067956	A2	20010117	EP 1999-916332	19990402
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
US 6218371	B1	20010417	US 1999-286098	19990402
JP 2002510644	T2	20020409	JP 2000-542030	19990402
US 2002064515	A1	20020530	US 2001-824468	20010402

PRIORITY APPLN. INFO.:
US 1998-80729P P 19980403
US 1999-286098 A3 19990402
WO 1999-US7335 W 19990402

OTHER SOURCE(S): MARPAT 131:285393

AB The present invention relates to **synergistic** combinations of immunostimulatory **CpG** oligonucleotides and immunopotentiating cytokines. The immunopotentiating cytokine is GM-CSF, interleukin 3,

interleukin 5, **interleukin 12**, interferon .gamma., TNF.alpha., Flt3 ligand or fusion protein comprising the cytokine and an antigen. The immunostimulatory **CpG** oligonucleotides and immunopotentiating cytokine are used together with antigen selected from the group consisting of a tumor antigen, microbial antigen or allergen. The antigen and the adjuvant compn. is useful for immunotherapy.

L18 ANSWER 1 OF 1 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE 1
ACCESSION NUMBER: 2001:13340 BIOSIS
DOCUMENT NUMBER: PREV200100013340
TITLE: Intranasal immunization of mice with **CpG** DNA induces strong systemic and mucosal responses that are influenced by other mucosal adjuvants and antigen distribution.
AUTHOR(S): McCluskie, Michael J.; Weeratna, Risini D.; Davis, Heather L. (1)
CORPORATE SOURCE: (1) Loeb Health Research Institute, Ottawa Hospital, 725 Parkdale Avenue, Ottawa, K1Y 4E9: hdavis@LRI.ca Canada
SOURCE: Molecular Medicine (New York), (October, 2000) Vol. 6, No. 10, pp. 867-877. print.
ISSN: 1076-1551.
DOCUMENT TYPE: Article
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Background: Synthetic oligodeoxynucleotides (ODN) containing immunostimulatory cytosine-guanine phosphate-linked dinucleotide (**CpG**) motifs are potent systemic and mucosal adjuvants in mice that have **synergistic** action with numerous other adjuvants, including alum and cholera toxin (CT). Herein, we evaluate **CpG** ODN with intranasal (IN) delivery of purified hepatitis B surface antigen (HBsAg), relative to and in combination with CT, Escherichia coli heat labile enterotoxin (LT), the B subunit of CT (CTB), and a nontoxic derivative of LT (LTK63). Materials and Methods: BALB/c mice were immunized by IN administration of HBsAg, alone or combined with CT, LT, CTB, or LTK63, and/or **CpG** ODN, or non-**CpG** control ODN. In addition, the effect of low-or high-volume administration was assessed, in order to target upper respiratory or entire respiratory tract, respectively. HBsAg-specific systemic (immunoglobulins: IgG, IgG1, IgG2a in plasma) and mucosal (IgA in fecal, lung, vaginal, saliva, and gut samples) humoral responses, as well as cell-mediated immune responses including T-cell proliferation and cytokines (interleukins: IL-4, **IL-5**; interferon: IFN-gamma) were evaluated. Results: **CpG** ODN, CT, and LT augmented anti-HBs titers equally, and more so than did CTB or LTK63. **CpG** ODN acted **synergistically** with CT and LT, but not CTB or LTK63 to enhance anti-HBs titers. Nevertheless, **CpG** ODN induced a more Thl-like response for all combinations, compared with the same formulation without **CpG**. Strength of induced systemic and mucosal immune responses was better with IN delivery of a large volume. A small volume required multiple administrations and higher doses of antigen and adjuvant for equal results. This suggests that delivery of antigen to the lung and/or diges-tive system is superior to delivery to the nasal cavity. Conclusions: Our results suggest that the synergy between **CpG** ODN and native toxins (CT, LT) may depend on their enzymatic activity and that the lack of synergy with nontoxic derivatives (LTB, LTK63) arises, since they do not have enzymatic activity. Because both CT and LT are too toxic for use in humans, it is possible that **CpG** ODN may be combined with bacterial toxin mutants that retain some enzymatic activity to optimize immune augmentation.

L20 ANSWER 1 OF 1 MEDLINE
ACCESSION NUMBER: 2001544514 MEDLINE
DOCUMENT NUMBER: 21475558 PubMed ID: 11592079
TITLE: Toll-like receptor expression reveals **CpG** DNA as a unique microbial stimulus for plasmacytoid dendritic cells which synergizes with CD40 ligand to induce high amounts of IL-12.
AUTHOR: Krug A; Towarowski A; Britsch S; Rothenfusser S; Hornung V; Bals R; Giese T; Engelmann H; Endres S; Krieg A M; Hartmann G
CORPORATE SOURCE: Department of Internal Medicine and Division of Clinical Pharmacology, University of Munich, Munich, Germany.
SOURCE: EUROPEAN JOURNAL OF IMMUNOLOGY, (2001 Oct) 31 (10) 3026-37. Journal code: 1273201. ISSN: 0014-2980.
PUB. COUNTRY: Germany: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200112
ENTRY DATE: Entered STN: 20011010
Last Updated on STN: 20020122
Entered Medline: 20011204

AB Human plasmacytoid dendritic cells (DC) (PDC, CD123+) and myeloid DC (MDC, CD11c+) may be able to discriminate between distinct classes of microbial molecules based on a different pattern of Toll-like receptor (TLR) expression. TLR1-TLR9 were examined in purified PDC and MDC. TLR9, which is critically involved in the recognition of **CpG** motifs in mice, was present in PDC but not in MDC. TLR4, which is required for the response to LPS, was selectively expressed on MDC. Consistent with TLR expression, PDC were susceptible to stimulation by **CpG** oligodeoxynucleotide (ODN) but not by LPS, while MDC responded to LPS but not to **CpG** ODN. In PDC, **CpG** ODN supported survival, activation (CD80, CD86, CD40, MHC class II), chemokine production (IL-8, IP-10) and maturation (CD83). CD40 ligand (CD40L) and **CpG** ODN synergized to activate PDC and to stimulate the production of IFN-alpha and IL-12 including bioactive IL-12 p70. Previous incubation of PDC with **IL-3** decreased the amount of **CpG**-induced IFN-alpha and shifted the cytokine response in favor of IL-12. **CpG** ODN-activated PDC showed an increased ability to stimulate proliferation of naive allogeneic CD4 T cells, butTh1 polarization of developing T cells required simultaneous activation of PDC by CD40 ligation and **CpG** ODN. **CpG** ODN-stimulated PDC expressed CCR7, which mediates homing to lymph nodes. In conclusion, our studies reveal that IL-12 p70 production by PDC is under strict control of two signals, an adequate exogenous microbial stimulus such as **CpG** ODN, and CD40L provided endogenously by activated T cells. Thus, **CpG** ODN acts as an enhancer of T cell help, while T cell-controlled restriction to foreign antigens is maintained.

L23 ANSWER 1 OF 9 MEDLINE DUPLICATE 1
ACCESSION NUMBER: 2001357881 MEDLINE
DOCUMENT NUMBER: 21311855 PubMed ID: 11418633
TITLE: Novel roles of **CpG** oligodeoxynucleotides as a leader for the sampling and presentation of **CpG** -tagged antigen by dendritic cells.
AUTHOR: Shirota H; Sano K; Hirasawa N; Terui T; Ohuchi K; Hattori T; Shirato K; Tamura G
CORPORATE SOURCE: First Department of Internal Medicine and Department of Dermatology, Tohoku University School of Medicine, Sendai, Japan.
SOURCE: JOURNAL OF IMMUNOLOGY, (2001 Jul 1) 167 (1) 66-74. Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200109
ENTRY DATE: Entered STN: 20010924
Last Updated on STN: 20010924
Entered Medline: 20010920

AB Oligodeoxynucleotides containing **CpG** motifs have been highlighted as potent Th1 activators. We previously reported that Ag and **CpG**, when conjugated together, **synergistically** promoted the Ag-specific Th1 development and inhibited the Th2-mediated airway eosinophilia. In this study, we examined the mechanisms underlying the **synergism** of the covalent conjugation. The **CpG**-OVA conjugate enhanced the Th1 activation and development. These characteristic features of the conjugate could not be ascribed to the polymerization of OVA, but mirrored the augmented binding of the **CpG**-tagged Ag to dendritic cells (DCs) in a **CpG**-guided manner, because phycobiliprotein, R-PE, conjugated to **CpG** stained a higher proportion of DCs with higher intensity than the mixture. R-PE fluorescence was emitted from cytoplasmic portions of the DCs, which simultaneously expressed costimulatory molecules and **IL-12**. The **CpG**-conjugated R-PE trafficking described above actually served as a potent Ag. These results indicate that **CpG** conjugated to Ag exhibit novel joint properties as promoters of Ag uptake and DC activators, thereby potentiating the ability of DCs to generate Th1 cells. The DNA-mediated promotion of Ag uptake would be advantageous for evoking host immune responses against invading microorganisms.

L23 ANSWER 2 OF 9 MEDLINE DUPLICATE 2
ACCESSION NUMBER: 2000281654 MEDLINE
DOCUMENT NUMBER: 20281654 PubMed ID: 10820244
TITLE: **CpG** oligonucleotides are potent adjuvants for the activation of autoreactive encephalitogenic T cells in vivo.
AUTHOR: Segal B M; Chang J T; Shevach E M
CORPORATE SOURCE: Laboratory of Immunology, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Bethesda, MD 20892, USA.
SOURCE: JOURNAL OF IMMUNOLOGY, (2000 Jun 1) 164 (11) 5683-8. Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals; AIDS
ENTRY MONTH: 200006
ENTRY DATE: Entered STN: 20000629
Last Updated on STN: 20000629
Entered Medline: 20000621

AB The mechanism of action of microbial adjuvants in promoting the differentiation of autoimmune effector cells remains to be elucidated. We

demonstrate that **CpG**-containing oligodeoxynucleotides (ODN) can completely substitute for heat-killed mycobacteria in the priming of encephalitogenic myelin-reactive T cells in vivo. The adjuvanticity of the **CpG** ODN was secondary to their direct ability to induce **IL-12** or to act **synergistically** with endogenous **IL-12** to promote Th1 differentiation and encephalitogenicity. T cells primed in the absence of **CpG** with Ag and IFA alone appeared to be in a transitional state and had not undergone differentiation along a conventional Th pathway. Unlike Th2 cells, they expressed low levels of the IL-12R beta 2 subunit and retained the ability to differentiate into encephalitogenic effectors when reactivated in vitro under Th1-polarizing conditions. These results support the use of **CpG** ODN as adjuvants but also suggest that they could potentially trigger autoimmune disease in a susceptible individual.

L23 ANSWER 3 OF 9 MEDLINE DUPLICATE 3
ACCESSION NUMBER: 2000281641 MEDLINE
DOCUMENT NUMBER: 20281641 PubMed ID: 10820231
TITLE: Regulation of murine airway eosinophilia and Th2 cells by antigen-conjugated **CpG** oligodeoxynucleotides as a novel antigen-specific immunomodulator.
AUTHOR: Shirota H; Sano K; Kikuchi T; Tamura G; Shirato K
CORPORATE SOURCE: First Department of Internal Medicine, Tohoku University School of Medicine, Sendai, Japan.
SOURCE: JOURNAL OF IMMUNOLOGY, (2000 Jun 1) 164 (11) 5575-82. Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals; AIDS
ENTRY MONTH: 200006
ENTRY DATE: Entered STN: 20000629
Last Updated on STN: 20000629
Entered Medline: 20000621

AB The characteristic features of bronchial asthma reflect the orchestrated activity of Th2 cells. Oligodeoxynucleotides containing **CpG** motifs (**CpG**) have recently been highlighted as an immunomodulator that biases toward a Th1-dominant phenotype. We have previously reported that intratracheal coadministration of **CpG** and allergen inhibited airway eosinophilia and hyperresponsiveness in a **synergistic** manner. To substantiate the **synergism** between **CpG** and Ag, we introduced a covalently linked conjugate between **CpG** and Ag and examined the efficacy on airway eosinophilia and Th2 cytokine production. We found that the conjugated form of **CpG** plus Ag was 100-fold more efficient in regulating airway eosinophilia than the unconjugated mixture. The inhibitory effects lasted for at least 2 mo. The inhibition of airway eosinophilia by the conjugate was Ag specific and associated with an improvement of the airway hyperresponsiveness and the unresponsiveness of the Ag-specific Th2 cells in the regional lymph nodes. The **CpG**-Ag conjugate was 100-fold more effective than the unconjugated mixture for inducing in vitro Th1 differentiation in an **IL-12**-dependent manner. Our data show that **CpG** conjugated to Ag can work as a novel Ag-specific immunomodulator and imply that inhalation of allergen-**CpG** conjugate could be a desensitization therapy for patients with bronchial asthma.

L23 ANSWER 4 OF 9 MEDLINE
ACCESSION NUMBER: 2002263872 MEDLINE
DOCUMENT NUMBER: 21990298 PubMed ID: 11994440
TITLE: IFN-alpha beta promote priming of antigen-specific CD8+ and CD4+ T lymphocytes by immunostimulatory DNA-based vaccines.
AUTHOR: Cho Hearn Jay; Hayashi Tomoko; Datta Sandip K; Takabayashi

Kenji; Van Uden John Henry; Horner Anthony; Corr Maripat; Raz Eyal

CORPORATE SOURCE: Division of Hematology/Medical Oncology, Department of Medicine, New York Presbyterian Hospital and Cornell Medical Center, 525 East 68th Street, New York, NY 10021.. hjc2001@med.cornell.edu

CONTRACT NUMBER: AI 40682 (NIAID)
AI 47078 (NIAID)
AR 44850 (NIAMS)

SOURCE: JOURNAL OF IMMUNOLOGY, (2002 May 15) 168 (10) 4907-13. Journal code: 2985117R. ISSN: 0022-1767.

PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200206
ENTRY DATE: Entered STN: 20020511
Last Updated on STN: 20020620
Entered Medline: 20020619

AB Immunostimulatory sequence (ISS) DNA containing unmethylated **CpG** dinucleotides stimulate NK and APC to secrete proinflammatory cytokines, including IFN-alphabeta and -gamma, TNF-alpha, and IL-6 and -12, and to express costimulatory surface molecules such as CD40, B7-1, and B7-2. Although ISS DNA has little direct effect on T cells by these criteria, immunization of wild-type mice with ISS DNA and OVA results in Ag-specific CTL and Th1-type T helper activity. This investigation examines the mechanisms by which ISS DNA primes CD8(+) and CD4(+) lymphocyte activities. In this report we demonstrate that ISS DNA regulates the expression of costimulatory molecules and TAP via a novel autocrine or paracrine IFN-alphabeta pathway. Coordinated regulation of B7 costimulation and TAP-dependent cross-presentation results in priming of Ag-specific CD8(+) CTL, whereas CD40, B7, and **IL-12** costimulation is required for priming of CD4(+) Th cells by ISS-based vaccines.

L23 ANSWER 5 OF 9 MEDLINE
ACCESSION NUMBER: 2002003622 MEDLINE
DOCUMENT NUMBER: 21623906 PubMed ID: 11751985
TITLE: Colony-stimulating factor-1 suppresses responses to **CpG** DNA and expression of toll-like receptor 9 but enhances responses to lipopolysaccharide in murine macrophages.

AUTHOR: Sweet Matthew J; Campbell Carol C; Sester David P; Xu Damo; McDonald Rebecca C; Stacey Katryn J; Hume David A; Liew Foo Y

CORPORATE SOURCE: Department of Immunology and Bacteriology, University of Glasgow, Glasgow, United Kingdom.. M.Sweet@imb.uq.edu.au

SOURCE: JOURNAL OF IMMUNOLOGY, (2002 Jan 1) 168 (1) 392-9. Journal code: 2985117R. ISSN: 0022-1767.

PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200201
ENTRY DATE: Entered STN: 20020102
Last Updated on STN: 20020125
Entered Medline: 20020111

AB During bacterial infections, the balance between resolution of infection and development of sepsis is dependent upon the macrophage response to bacterial products. We show that priming of murine bone marrow-derived macrophages (BMMs) with CSF-1 differentially regulates the response to two such stimuli, LPS and immunostimulatory (**CpG**) DNA. CSF-1 pretreatment enhanced IL-6, **IL-12**, and TNF-alpha production in response to LPS but suppressed the same response to

CpG DNA. CSF-1 also regulated cytokine gene expression in response to CpG DNA and LPS; CpG DNA-induced IL-12 p40, IL-12 p35, and TNF-alpha mRNAs were all suppressed by CSF-1 pretreatment. CSF-1 pretreatment enhanced LPS-induced IL-12 p40 mRNA but not TNF-alpha and IL-12 p35 mRNAs, suggesting that part of the priming effect is posttranscriptional. CSF-1 pretreatment also suppressed CpG DNA-induced nuclear translocation of NF-kappaB and phosphorylation of the mitogen-activated protein kinases p38 and extracellular signal-related kinases-1/2 in BMMs, indicating that early events in CpG DNA signaling were regulated by CSF-1. Expression of Toll-like receptor (TLR)9, which is necessary for responses to CpG DNA, was markedly suppressed by CSF-1 in both BMMs and thioglycolate-elicited peritoneal macrophages. CSF-1 also down-regulated expression of TLR1, TLR2, and TLR6, but not the LPS receptor, TLR4, or TLR5. Hence, CSF-1 may regulate host responses to pathogens through modulation of TLR expression. Furthermore, these results suggest that CSF-1 and CSF-1R antagonists may enhance the efficacy of CpG DNA in vivo.

L23 ANSWER 6 OF 9 MEDLINE
ACCESSION NUMBER: 2001544765 MEDLINE
DOCUMENT NUMBER: 21475889 PubMed ID: 11591791
TITLE: Intracisternally localized bacterial DNA containing CpG motifs induces meningitis.
AUTHOR: Deng G M; Liu Z Q; Tarkowski A
CORPORATE SOURCE: Department of Rheumatology, Goteborg University, Goteborg, Sweden.. guo-min@rheuma.gu.se
SOURCE: JOURNAL OF IMMUNOLOGY, (2001 Oct 15) 167 (8) 4616-26. Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200112
ENTRY DATE: Entered STN: 20011010
Last Updated on STN: 20020122
Entered Medline: 20011207

AB Unmethylated CpG motifs are frequently found in bacterial DNA, and have recently been shown to exert immunostimulatory effects on leukocytes. Since bacterial infections in the CNS will lead to local release of prokaryotic DNA, we wanted to investigate whether such an event might trigger meningitis. To that end, we have intracisternally injected mice and rats with bacterial DNA and oligonucleotides containing CpG motifs. Histopathological signs of meningitis were evident within 12 h and lasted for at least 14 days, and were characterized by an influx of monocytic, Mac-3(+) cells and by a lack of T lymphocytes. To study the mechanisms whereby unmethylated CpG DNA gives rise to meningitis, we deleted the monocyte/macrophage population leading to abrogation of brain inflammation. Also, interaction with NF-kappaB using antisense technology led to down-regulation of proinflammatory cytokine production and frequency of meningitis. Furthermore, specific interactions with vascular selectin expression and inhibition of NO synthase led to a significant amelioration of meningitis, altogether indicating that this condition is dependent on macrophages and their products. In contrast, neutrophils, NK cells, T/B lymphocytes, IL-12, and complement system were not instrumental in meningitis triggered by bacterial DNA containing CpG motifs. This study proves that bacterial DNA containing unmethylated CpG motifs induces meningitis, and indicates that this condition is mediated in vivo by activated macrophages.

L23 ANSWER 7 OF 9 MEDLINE
ACCESSION NUMBER: 2001544514 MEDLINE

DOCUMENT NUMBER: 21475558 PubMed ID: 11592079
TITLE: Toll-like receptor expression reveals **CpG** DNA as a unique microbial stimulus for plasmacytoid dendritic cells which synergizes with CD40 ligand to induce high amounts of **IL-12**.
AUTHOR: Krug A; Towarowski A; Britsch S; Rothenfusser S; Hornung V; Bals R; Giese T; Engelmann H; Endres S; Krieg A M; Hartmann G
CORPORATE SOURCE: Department of Internal Medicine and Division of Clinical Pharmacology, University of Munich, Munich, Germany.
SOURCE: EUROPEAN JOURNAL OF IMMUNOLOGY, (2001 Oct) 31 (10) 3026-37. Journal code: 1273201. ISSN: 0014-2980.
PUB. COUNTRY: Germany: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200112
ENTRY DATE: Entered STN: 20011010
Last Updated on STN: 20020122
Entered Medline: 20011204

AB Human plasmacytoid dendritic cells (DC) (PDC, CD123+) and myeloid DC (MDC, CD11c+) may be able to discriminate between distinct classes of microbial molecules based on a different pattern of Toll-like receptor (TLR) expression. TLR1-TLR9 were examined in purified PDC and MDC. TLR9, which is critically involved in the recognition of **CpG** motifs in mice, was present in PDC but not in MDC. TLR4, which is required for the response to LPS, was selectively expressed on MDC. Consistent with TLR expression, PDC were susceptible to stimulation by **CpG** oligodeoxynucleotide (ODN) but not by LPS, while MDC responded to LPS but not to **CpG** ODN. In PDC, **CpG** ODN supported survival, activation (CD80, CD86, CD40, MHC class II), chemokine production (IL-8, IP-10) and maturation (CD83). CD40 ligand (CD40L) and **CpG** ODN synergized to activate PDC and to stimulate the production of IFN-alpha and **IL-12** including bioactive **IL-12** p70. Previous incubation of PDC with IL-3 decreased the amount of **CpG**-induced IFN-alpha and shifted the cytokine response in favor of **IL-12**. **CpG** ODN-activated PDC showed an increased ability to stimulate proliferation of naive allogeneic CD4 T cells, butTh1 polarization of developing T cells required simultaneous activation of PDC by CD40 ligation and **CpG** ODN. **CpG** ODN-stimulated PDC expressed CCR7, which mediates homing to lymph nodes. In conclusion, our studies reveal that **IL-12** p70 production by PDC is under strict control of two signals, an adequate exogenous microbial stimulus such as **CpG** ODN, and CD40L provided endogenously by activated T cells. Thus, **CpG** ODN acts as an enhancer of T cell help, while T cell-controlled restriction to foreign antigens is maintained.

L23 ANSWER 8 OF 9 MEDLINE
ACCESSION NUMBER: 2000155542 MEDLINE
DOCUMENT NUMBER: 20155542 PubMed ID: 10693875
TITLE: The features of arthritis induced by **CpG** motifs in bacterial DNA.
AUTHOR: Deng G M; Tarkowski A
CORPORATE SOURCE: Department of Rheumatology, University of Goteborg, Sweden.
SOURCE: ARTHRITIS AND RHEUMATISM, (2000 Feb) 43 (2) 356-64. Journal code: 0370605. ISSN: 0004-3591.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200003
ENTRY DATE: Entered STN: 20000320
Last Updated on STN: 20000320

Entered Medline: 20000309

AB OBJECTIVE: To investigate the features of arthritis induced by bacterial DNA that contain **CpG** motifs. METHODS: Bacterial DNA originating from Escherichia coli and Staphylococcus aureus or synthetic oligonucleotides containing **CpG** motifs were injected directly into knee joints of mice. Histopathologic joint damage, antibody levels, cytokine levels, and synovial messenger RNA (mRNA) expression of cytokines and chemokines were assessed. RESULTS: Histopathologic signs of arthritis were evident within 2 hours and lasted for at least 3 weeks. Nonmethylated **CpG** motifs were responsible for the induction of arthritis since oligonucleotides containing these motifs triggered arthritis, whereas methylation of these nucleotides abrogated the inflammatory response. Arthritis was characterized by an influx of monocytic, Mac-1+ cells and by a scarcity of T lymphocytes. The disease was characterized locally by mRNA expression of macrophage-derived cytokines (tumor necrosis factor alpha, interleukin-12 [**IL-12**], IL-1beta) and chemokines (monocyte chemoattractant protein 1, RANTES) in arthritic joints. Systemically, the arthritis was characterized by increased levels of circulating IL-6 and immunoglobulins. CONCLUSION: These findings demonstrate that bacterial DNA that contain nonmethylated **CpG** motifs induces arthritis, suggesting an important pathogenic role for bacterial DNA in septic arthritis.

L23 ANSWER 9 OF 9 MEDLINE
ACCESSION NUMBER: 1998451440 MEDLINE
DOCUMENT NUMBER: 98451440 PubMed ID: 9780160
TITLE: Cyclosporin A enhances **IL-12** production by **CpG** motifs in bacterial DNA and synthetic oligodeoxynucleotides.
AUTHOR: Redford T W; Yi A K; Ward C T; Krieg A M
CORPORATE SOURCE: University of Iowa College of Pharmacy, Iowa City 52242, USA.
CONTRACT NUMBER: DK25295 (NIDDK)
P01CA665078 (NCI)
R29-AR42556 (NIAMS)
SOURCE: JOURNAL OF IMMUNOLOGY, (1998 Oct 15) 161 (8) 3930-5.
Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199811
ENTRY DATE: Entered STN: 19990106
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AB Certain sequences of nucleotides (**CpG** motifs) in bacterial DNA or synthetic oligonucleotides (**CpG** DNA) promote the production of proinflammatory cytokines, including TNF-alpha, IFN-gamma, IL-6, and **IL-12**. Here we demonstrate that the immunosuppressant cyclosporin A (CsA) unexpectedly enhanced **CpG** DNA-induced **IL-12** production in murine splenocytes. CsA did not inhibit **CpG** DNA-induced TNF-alpha or IL-6 production, but decreased the production of IFN-gamma by **CpG** DNA. Upon examining mechanisms by which CsA increases **IL-12** production, we found that **CpG** DNA can also induce IL-10 production in B cells and that this production was sensitive to CsA. IL-10 has anti-inflammatory effects and can reduce the production of **IL-12**. To determine the possible role of CsA-modulated IL-10 production in mediating the increased **IL-12** levels, splenocytes from IL-10 gene-disrupted mice (IL-10 -/-) and splenocytes cultured in anti-IL-10 Ab were studied. **CpG** DNA-stimulated IL-10 (-/-) splenocytes demonstrated no increase in **IL-12** levels in the presence of CsA. Anti-IL-10 Ab treatment of normal splenocytes increased the magnitude of **CpG** DNA-induced **IL-12**

production to that seen with CsA. These results suggest that CpG DNA induces CsA-sensitive IL-10 production in B cells and that IL-10 acts as a negative feedback regulator of CpG DNA-induced IL-12 production.